AD-A174 613 PARDAXIN'S ACTION IN SHARK(U) NEW YORK AQUARIUM BROOKLYN OSBORN LABS OF MARINE SCIENCES N PRIMOR ET AL 24 OCT 86 N00014-82-C-0435 1/1

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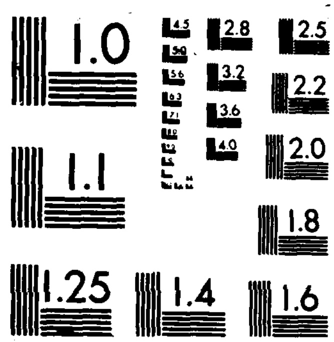

CROCOPY RESOLUTION TEST CHART

new york aquarium

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October 24, 1986

AD-A174 613

Dr. Eli D. Schmell
Code 1141MB
Office of Naval Research
800 North Quincy St.
Arlington, VA 22217-5000

Dear Dr. Schmell:

Enclosed please find a Progress Report. In addition, I am enclosing a paper which is being accepted for publication in J.Biol. Chem.. It is expected to be published in February-March, 1987. This is a major paper on the chemistry and the mechanism of ion-channel formation of pardaxin.

Sincerely,

Naftali Primor

Naftali Primor

NP:ms
Enc.

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Title Project: Pardaxin's action in shark

Naftali Primor, Philip Lazarovici

Osborn Laboratories of Marine Sciences

Brooklyn, New York 11224

Contract: N 00014-82-C-0435

Objectives:

Structure and mode of action of the shark repellent pardaxin

Abstract

1. Amino Acid analysis

The Amino acid composition of pardaxins I and II purified from *P. marmoratus* secretion is listed.

Amino acid	PXI	PXII
Asp	4.22 (4)	3.79 (4)
Thr	1.23 (1)	0.92 (1)
Ser	3.78 (4)	3.75 (4)
Glu	2.98 (3)	2.71 (3)
Pro	0.76 (1)	1.05 (1)
Gly	6.33 (6)	4.23 (4)
Ala	2.82 (3)	2.83 (3)
Val	1.77 (2)	1.13 (1)
Ile	1.93 (2)	1.85 (2)
Leu	2.25 (2)	0.62 (1)
Tyr	0	0
Phe	2.27 (2)	1.80 (2)
His	0.95 (1)	0.94 (1)
Lys	1.91 (2)	1.23 (1)
Arg	0.33 (0)	0.23 (0)
Trp	0	0
Cys	1.32 (2)	1.51 (2)

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Met	0	1.91 (1)
Total Residues	35	31
Molecular weight	3529	3188
Total hydrophylic(3	48.5%	45.2%
Total hydrophobic(4	51.5%	54.8%

2. Sequence determination of pardaxin I

Pardaxins were analyzed by automated Edman degradation in a Beckman sequencer 890 C. The degradation was carried out using 0.25 M Quadrol, 5% phenylisothiocyanate with single cleavage. Conversion of teh phenylthiazo derivatives was achieved with 1 N HCl for 10 min at 80°C. The phenylthiohydantoin (PTH) derivatives were identified by high pressure liquid chromatography.

The amino acid sequence of the NH_2-terminal of pardaxin I:

Cycle No.	Amino acid
1	Gly
2	Phe
3	Phe
4	Ala
5	Leu
6	Ile
7	Pro
8	Gly
9	Ile
10	Glu

Contd

3. Binding of Pardaxin to shark gill membranes is accomplished

Membranes from a blue shark (Prionace) gills were used. A sucrose gradient was used for membrane purification. Pardaxin was iodinated using the method of Bolton-Hunter reagent (as described in the paper JBC, In press.). We are in the process of evaluating this data.

Plans for next year:

1. Synthesis of N-terminal.
2. Test for activities of the synthetized N terminal:
   a. hydrophobicity
   b. Compative replacement with pardaxin using membranes
   c. cytotoxicity

3. Binding of the iodinated pardaxin to shark gill membranes

Published work on the subject (1986)

Primor, N. Action on ileal smooth muscle of synthetic detergents and pardaxin. Gen. Pharmocal. 17, 413-418 (1986).

Lazarovici, P., Primor, N. and Loew, L. Purification and pore forming activity of two hydrophodic polypeptides from the Red Sea Moses sole. J. Biol. Chem. In press.

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